

Vegetative propagation of *Litsea monopetala*, a wild tropical medicinal plant: Effects of indole-3-butyric acid (IBA) on stem cuttings

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Abstract: In this study we investigated the rooting ability and the growth performance of juvenile single-node leafy stem cuttings of *Litsea monopetala* (Roxb) Pers. collected from two mature mother trees preserved in the hill forest of Chittagong district, Bangladesh. The rooting ability of cuttings was studied under 0%, 0.1%, 0.2% and 0.4% indole-3-butyric acid (IBA) treatments. Significantly better rooting response ($p \leq 0.05$) was observed with 0.1% IBA compared to control (0% IBA). The mean number of roots and the length of the longest root of cuttings in different treatments showed no significant difference ($p \leq 0.05$). After transfer into polythene bags from non-mist propagator, rooted cuttings treated with 0%, 0.1% and 0.2% IBA demonstrated the highest ($100 \pm 0.00\%$) survival capacity. The mean number of shoots developed in cuttings in the polythene bags in first three weeks varied significantly ($p \leq 0.05$) among the treatments. Effects of three fertilizer treatments, viz. T0 (no fertilizer), T1 (10g Urea, 20g TSP, 10g MOP dissolved in 1 L water) and T2 (10g Urea, 20g TSP, 10g MOP dissolved in 2 L water) on initial growth of

stecklings were also measured over a 90-days period. The increment of leaf area of stecklings was significantly higher ($p \leq 0.05$) under T0 compared with that under T1 and T2 while the increment of stem length, collar diameter and root biomass varied insignificantly among different fertilizer treatments. The results suggest that rooting juvenile single-node leafy stem cuttings could be an effective mean of regenerating *L. monopetala*. The application of 0.1% IBA concentration is recommended for rooting of juvenile leafy stem cuttings and application of fertilizer appeared unnecessary for the subsequent growth of stecklings in polythene bags.

Keywords: IBA; *Litsea monopetala*; non-mist propagator; rooting; stem cutting

Introduction

Litsea monopetala (Roxb) Pers., [common names - Menda, Meda, Bara, kukur-chita, Boi-bet, Bol-bek, Akarma, Lalkhori, Huoria] of the family Lauraceae is a medium-sized species, about 12 m high with stalked, alternate and oblong to ovate shaped leaves (Manandhar 2002). It is naturally found in the sub-Himalayan tract - Nepal, India, Bangladesh, and Burma (Troup 1986). The bark is rough, with deep irregular cracks, blaze buff with light steaks, not greasy and blade 10–20 cm long (Chaudhury 1993). This is a valuable medicinal tree species - barks, leaves and roots are used by traditional healers, rural communities and pharmaceutical companies for remedies of diseases such as gonorrhea, skin diseases, boil etc (Troup 1986; Baul 2006). The leaves are used as a topical medicine for the treatment of arthritis (Puhua et al. 2008).

Though the propagation of this species is possible by seeds, the lack of abundant seeds made germination - the uncertain and time consuming option. This is caused as the local populations harvest the full tree or cut the large branches to address their fuel or fodder needs without ecological concerns mainly due to ignorance. Indiscriminate harvesting is dangerously reducing seed production, inducing pathogenic infections to standing

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healthy trees and causing physiological stresses resulting in gradual erosion of the natural populations and leading to the extinction of this tree species (Baul 2006). Therefore, for the preservation and domestication of *L. monopetala* alongwith taping its medicinal values, the establishment of vegetative methods of propagation such as stem cutting is of great importance.

In some parts of Asia, agroforestry is now being widely practiced and the domestication of this species on farm agroforestry through vegetative propagation would be advantageous. Clonal propagation techniques offer the opportunity to produce a reliable and adequate supply of superior planting stock locally, timely and quickly. Many tropical tree species have been successfully propagated vegetatively by stem cuttings (Leakey et al. 2006; Tchoundjeu et al. 2006). This approach is considered the ideal method for rapid multiplication by maintaining certain desirable traits of a threatened species (Hartmann et al. 2002; Tchoundjeu et al. 2004). Since species differs in their rooting requirements (Leakey et al. 1994) and rooting percentage differs with different concentrations of individual auxins (Leakey et al. 1990; Tchoundjeu et al. 2002) and no serious attempt was being observed to propagate *L. monopetala*, it is crucial to determine the appropriate treatment for optimum rooting of this species for vegetative propagation.

With an aim to assist preserving this species from economic and biodiversity point of views, this experiment was designed to investigate the possibility of vegetative propagation of *L. monopetala* by investigating the effects of different IBA concentrations on the rooting ability of juvenile leafy cuttings from mother tree plants. The effects of various fertilizer treatments on the initial growth of the rooted cuttings after transferring to the poly bags was also investigated in order to determine appropriate maintenance of stocklings produced by vegetative means. The results have shown promising sign of the amenability of the species to vegetative propagation through stem cutting with relative ease.

Materials and methods

Non-mist propagator construction

The study on vegetative propagation was carried out in a low-cost non-mist propagator placed under the nursery shed of the Institute of Forestry and Environmental Sciences, University of Chittagong (23°36'30"N and 90°21'03"E), Bangladesh. The non-mist propagator was constructed following the design described by Leakey (1990) and Leakey et al. (1990) modified by Kamaluddin (1996). The propagator was made up of a wooden frame (length: 1.8 m; width: 1 m; height: 60 cm at one end and 45 cm at the other end) wrapped with a single sheet of transparent polythene such that the base is completely watertight and the lid is airtight. The frame was covered tightly with a single polythene sheet and the closely fitting lid. The polythene base of the propagator was covered with a 10 cm thick layer of moist coarse sand mixed with successive layers of fine gravels and small

stones. This layer supports rooting media kept in perforated plastic trays (10"×10"×4"). The propagator was kept under bamboo made shed to avoid excessive heat accumulation. Mean maximum and minimum temperature within the propagator during rooting was maintained at 24 and 33°C, respectively. The propagator was opened briefly in the morning and in the late afternoon to facilitate gas exchange. Whenever the propagator lid was opened for observation, a fine jet of water spraying was applied to cuttings to maintain a low vapor pressure deficit inside the propagator. This resulted in a permanently humid environment throughout the propagation period.

Cutting materials source and preparation

Two mature mother trees of 7–8 m height with superior phenotypic characteristics were selected from the hill forest of Chittagong district, Bangladesh. Single-node cuttings, 8–10 cm in length with one or two leaves trimmed with scissors up to 50% of total leaf area were harvested from selected juvenile shoots of demarcated mother trees. Cuttings were enclosed in polythene bags to check moisture loss and were transported quickly to the propagation site at the Institute of Forestry and Environmental Sciences, University of Chittagong. The cuttings were inserted obliquely into a standard rooting medium comprising of a mixture of coarse sand and fine gravels (diameter: 0.1–0.3 cm) treated with fungicide (Diathane M45) three days ahead of the experiment. The rooting medium was chosen based on previous works conducted on other tropical tree species. To account for the variability in rooting ability attributable to genetic differences, the same number of cuttings (120 cuttings) from each mother tree was used in each treatment.

IBA treatment, experimental design and performance of rooting of stem cuttings

A stock solution of 3.2% (v/v) indole-3-butyric acid (IBA) was prepared by dissolving the hormone in a 1:1 mixture of absolute ethanol and methanol (Negash 2003; Ofori et al. 1996). From the stock solution, by serial dilution in distilled water, 0.1, 0.2 and 0.4% IBA solutions were then prepared. Before inserting into the rooting medium, 10 µl of the IBA solution was applied to the base of each cutting using a micro-pipette, thus providing 10, 20 and 40 µg IBA per cutting for the 0.1%, 0.2% and 0.4% IBA, respectively. The control comprised of a comparable number of cuttings (60 cuttings) treated only with distilled water i.e., 0% IBA concentration.

The whole experiment was set up in randomized blocks, with each treatment replicated three times. Ten single-node stem cuttings collected from each stock plant were used per replicate to give a total of 240 cuttings (n = 240; 4 IBA concentrations × 2 mother trees × 10 cuttings × 3 replicates). Assessments of rooting success were done weekly after the first two weeks of cutting insertion into rooting medium. A cutting was considered as rooted when it had one or more roots exceeding 1 cm; the number and length of roots were also recorded.

Survival of rooted stem cuttings in polythene bags

After treatments cuttings were potted into 6"× 4" polythene bags containing a 3:1 mixture of forest soil and cow dung and they were arranged according to treatment under nursery shed for 3 weeks for their initial establishment. During this period, observation and recording were done to identify *alive* and *dead* rooted cuttings in polythene bags. Cuttings were defined as *dead* when severely rotted. Also numbers of new shoots of each steckling were recorded. Watering once a day was carried out during this period.

Fertilizer treatment, experimental design and performance of the established rooted cuttings

Established rooted cuttings (stecklings) in polythene bags were then kept in a designed nursery bed for another experiment of a 90-day period to determine the effects of different fertilizer treatments on their initial growth. Mixtures of Urea, Triple super phosphate (TSP) and Muriate of potash (MOP) were used in fertilizer treatments at different ratios. A total of 90 healthy stecklings of almost similar size were selected for two fertilizer treatments viz. T1 (10 g of Urea + 20 g of Triple super phosphate +10 g of Muriate of potash dissolved in 1 L of water) and T2 (10 g of urea + 20 g of Triple super phosphate + 10 g of Muriate of potash dissolved in 2 L of water) and a control, T0 (no fertilizer). All the stecklings were equally watered once a day throughout the observation period. The experiment was designed in randomized blocks and replicated 3 times ($n = 90$; 3 fertilizer treatments \times 10 stecklings \times 3 replicates). Stem length (measured by a measuring scale), collar diameter (measured by a slide calipers) and areas of growing leaves (lengths of leaves were measured by a measuring scale and those lengths were converted into areas by adopting dot grid method) of the stecklings under study were measured at a regular interval of 10 days. For root biomass measurement, three stecklings of each treatment were harvested at an interval of 45 days from day 0 to days 90. After measuring fresh weight, roots then were dried in an oven under 80°C temperature for 48 hours and their dry weight was measured.

Data analysis

All the statistical analyses were done by using Microsoft Excel 2007 and Statistical Package for Social Sciences (SPSS) v 16.0. The analysis of variance (ANOVA) procedures were used to test for significant effect of treatments, followed by Duncan's Multiple Range Test (DMRT) for comparisons of different means of different treatments. After collecting data - rooting percentage, mean number of root, mean longest length of rooted stem cuttings, survival percentage, mean number of shoot of established rooted cuttings after transfer in to the polythene bags and initial growth performance in terms of stem, collar diameter elongation, expansion of leaf and root biomass production of stecklings were calculated and analyzed.

Results

Effects of rooting hormone (IBA) on rooting ability

In the cuttings, rooting was initiated between 14th and 15th weeks for all the treatments and control. However, rooting formation in the cuttings treated with 0.1% was the quickest and the final rooting response was also significantly different ($p \leq 0.05$) among the four treatments. The highest percentage of rooting were observed in cuttings treated with 0.1% IBA followed by those treated with 0.2% IBA, 0.4% IBA and 0% IBA (Fig. 1). This indicates that the species is more responsive to lower levels of IBA concentration (0.1% IBA) for better rooting, after this level the rooting of cuttings seems to be less responsive to higher IBA concentrations. It is worth mentioning that 0.1% IBA treatment almost doubled the rooting response of cuttings. Though higher concentrations of IBA reduced rooting percentages, yet they were significantly above the control.

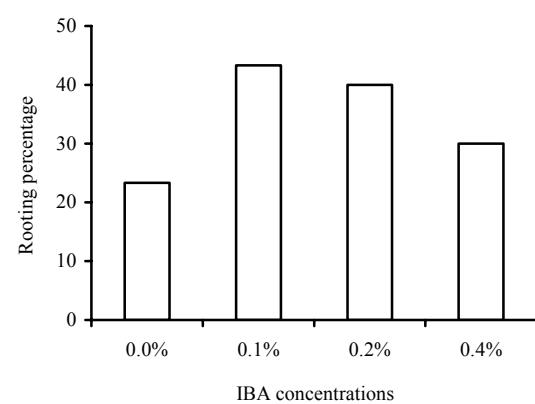


Fig. 1 Effects of various IBA concentrations on rooting ability of leafy stem cuttings of *Litsea monopetala*

The mean number of roots and mean longest root length per rooted cutting in this experiment did not change significantly ($p \leq 0.05$) among the four treatments. Mean number of roots per rooted cutting was found 2.43 ± 0.61 for the control, 2.46 ± 0.33 for 0.1%, 2.83 ± 0.55 for 0.2% and 2.11 ± 0.42 for 0.4% concentrations of IBA (Fig. 2). The length of the longest root indicated increment of root at different levels of IBA and control. The mean length of longest root was 4.82 ± 1.06 cm in cuttings treated with 0.1% IBA followed by 2.73 ± 0.78 cm, 2.72 ± 0.53 cm and 2.54 ± 0.70 cm for 0.4%, 0.2% and 0% levels of IBA, respectively (Fig. 3). At low concentration, IBA seems to have a positive role on increasing the root length but at higher concentrations, IBA does not promote root growth. From these results it can be inferred that the optimal dose of IBA, in this case 0.1% IBA, has a positive role on the morphology of the roots produced.

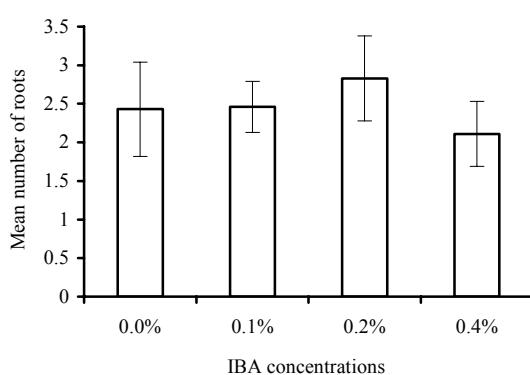


Fig. 2 Effects of various IBA concentrations on number of roots per cutting of *Litsea monopetala*. Bars represent \pm SE with $n = 60$ for each concentration of IBA

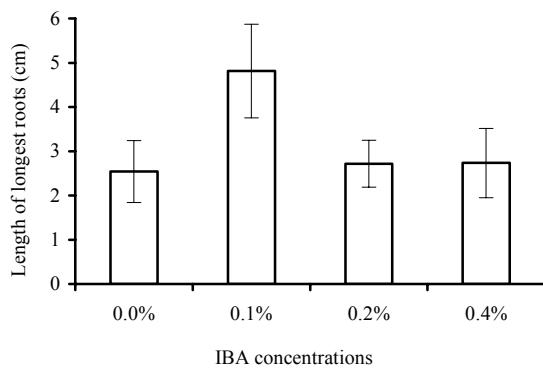


Fig. 3 Effects of various IBA concentrations on mean length of the longest roots of leafy stem cuttings of *Litsea monopetala*. Bars represent \pm SE

Survival of rooted cuttings after transfer to polythene bags

The overall survival of rooted cuttings after transferring them from non-mist propagator to polythene bags was high. Rooted cuttings treated with 0.1% IBA, 0.2% IBA and control demonstrated an highest survival capacity ($100 \pm 0.00\%$) whereas rooted cuttings treated with 0.4% IBA showed the lowest ($77.77 \pm 3.50\%$) survival values during an establishment period of 3 weeks in polythene bags (Fig. 4). The reduction in survival percentage at high IBA concentration was interesting. There was significant difference ($p \leq 0.05$) in the values of mean shoots number in the stecklings among four treatments. The mean number of shoots developed in the stecklings treated with 0.1% IBA was the highest (2.92 ± 0.18) followed by those of the stecklings treated with 0.4% IBA (2.44 ± 0.24), 0.2% IBA (2.25 ± 0.13) and 0% IBA (1.43 ± 0.20) during establishment periods in polythene bags (Fig. 5). Lower concentration of IBA (0.1%) was the most effective treatment to trigger shoots number of established rooted cuttings and higher concentrations of IBA was more effective than the control as well. It suggests that IBA has positive impact on shoot

development.

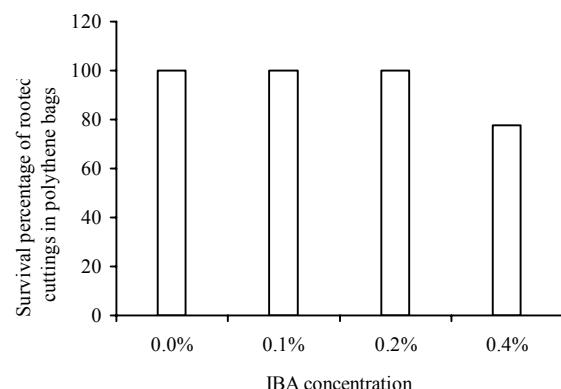


Fig. 4 Survival percentage of rooted cuttings of *Litsea monopetala* treated with different IBA concentrations in polythene bags

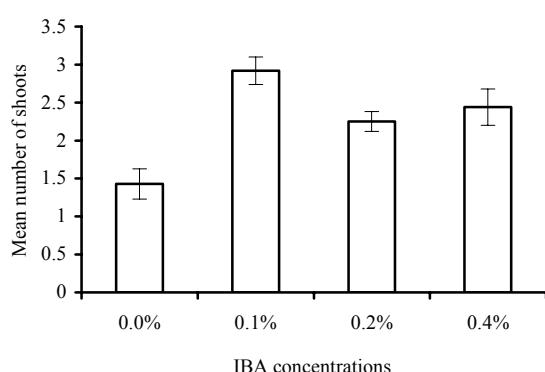


Fig. 5 Mean number of shoots of rooted cutting of *Litsea monopetala* treated with various IBA concentrations in polythene bags. Bars represent \pm SE

Initial growth performance of (stecklings) under different fertilizer treatments

Stem length of the stecklings in polythene bags was found to increase steadily throughout 90 days for fertilizer treatments T0 and T1 although increment of stecklings under T2 treatment was also steady after 20 days. In an expected way, stem length reached the highest values in the stecklings grown without fertilizer (control) followed by those treated with T2 and T1 throughout the observation period. However, this difference was not statistically significant proven ($p \leq 0.05$, Fig. 6).

Collar diameter increment of the stecklings in polythene bags was steady throughout 90 days for three treatments. Increment of collar diameter was the maximum in the stecklings under T1 compared to T0 and T2. After 20 days, T0 exhibited slightly better performance than T2 (Fig. 7). But no significant variation ($p \leq 0.05$) in collar diameter increment among fertilizer treatments was observed.

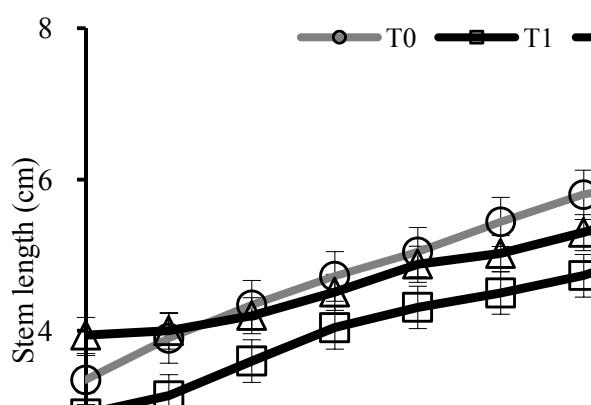


Fig. 6 Effects of different fertilizer treatments on stem length increment of established rooted cuttings (stecklings) of *Litsea monopetala*. T0 means No fertilizer, T1, Fertilizer treatment 1 and T2, Fertilizer treatment 2. Bars represent \pm SE with $n = 30$ for each fertilizer treatment.

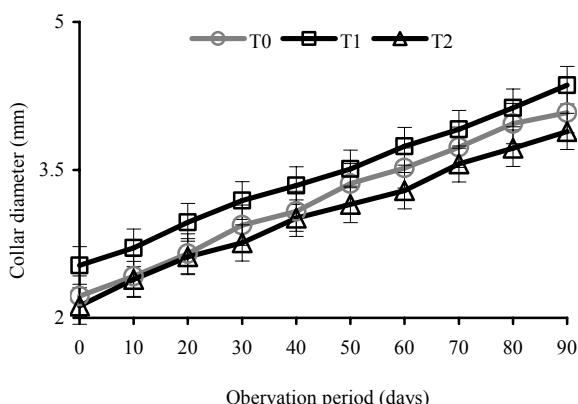


Fig. 7 Effects of different fertilizer treatments on collar diameter growth of established rooted cuttings (stecklings) of *Litsea monopetala*. T0 is no fertilizer, T1 is fertilizer treatment 1, and T2 is fertilizer treatment 2. Bars represent \pm SE with $n = 30$ for each fertilizer treatment.

There were increase of leaf areas of the stecklings in polythene bags throughout 90 days for the treatments, T1 and T2 and the control, T0. Leaf area was the significantly highest ($p \leq 0.05$) in the case of T0 stecklings among the three fertilizer treatments throughout the observation period (Fig. 8).

Root biomass on the basis of dry weight of the stecklings increased gradually over 90 days for three treatments. The increase of root biomass under treatment T1 was initially the highest followed by that under treatments T0 and T2 respectively but it was exceeded by that under treatment T0 after 45 days. At 45th day the root biomass of the stecklings under treatments T1 and T0 showed no significant difference while the same happened at 90th day under treatment T1 and T2 (Fig. 9). These results are indicative of the ability of vegetatively produced stacklings to utilize soil nutrients without the need for supplementary fertilizer treatments which is positive in the sense that the stacklings will require less cost for their subsequent maintenance and

establishment.

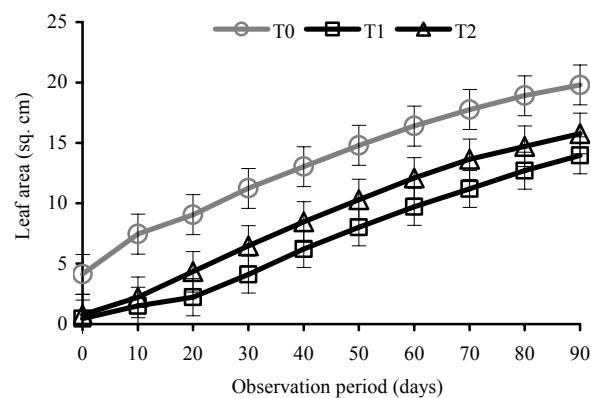


Fig. 8 Effects of different fertilizer treatments on leaf area expansion of established rooted cuttings (stecklings) of *Litsea monopetala*. T0, No fertilizer, T1, Fertilizer treatment 1, and T2, Fertilizer treatment 2. Bars represent \pm SE with $n = 30$ for each fertilizer treatment.

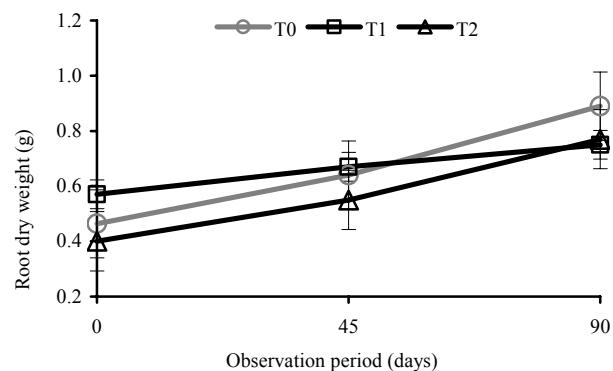


Fig. 9 Effects of different fertilizer treatments on root biomass increase of established rooted cuttings (stecklings) of *Litsea monopetala*. T0 (No fertilizer), T1 is Fertilizer treatment 1 and T2, Fertilizer treatment 2. Bars represent \pm SE with $n = 30$ for each fertilizer treatment.

Discussions

The adventitious root formation is regulated by optimal environmental conditions (cutting origin and environment) to achieve good rooting; optimal morphological and physiological condition of the cutting material, which in turn, affected by stockplant physiology and management (Dick et al. 2004; Leakey 2004). Consequently, genetic potential, as well as propagation environment, post-severance treatment, cutting origin and environment, stockplant physiology and management, have been reported to influence rooting (Hartmann et al. 2002). We can postulate that differences in rooting ability between clones could be due to genetic differences in cutting morphology and/or physiology (Leakey 2004) and naturally occurring auxin IAA and synthetic auxins, which affect metabolic changes such as starch, protein in cutting ultimately may have effect on adventitious root formation of woody stem cuttings (Hussen and Pal 2007a).

The application of auxins to induce root formation on leafy

stem cuttings is widely recognized (Leakey et al. 1990; Tchoundjeu and Leakey 2001; Husen 2003; Husen et al. 2003; Husen and Pal 2006, 2007a). Although rooting hormone named indole-3-butyric acid (IBA) has been used for rooting of various tropical tree species (Leakey et al. 1992; Ofori et al. 1996; Mese'n et al. 1997; Berhe and Negash 1998; Negash 2002; Tchoundjeu et al. 2002, 2004; Gateable' and Pastor 2006; Husen and Pal 2007a; Opuni-Frimpong et al. 2008; Amri et al. 2009), the different concentration of IBA applied leading to rooting response varied from species to species. Results of previous studies indicated different optimal IBA concentrations for suitable root responses: 20 µg per cutting (0.2% IBA) for *Shorea leprosula* (Aminah et al. 1995), 40 µg per cutting (0.4% IBA) for *Stereospermum suaveolens* (Baul et al. 2009), and 100–200 µg per cutting for *Prunus africana* (Tchoundjeu et al. 2002). *L. monopetala*, in this study, rooted better at an IBA concentration of 0.1% (10 µg per cutting) on leafy stem cuttings derived from mature mother trees while some published results for example- Mese'n et al. (1997) working on *Cordia alliodora* and Opuni-Frimpong et al. (2008) working on *Khaya anthotheca* and *Khaya ivorensis* showed a reverse trend with the percentage of rooting increasing with increasing IBA concentrations. By contrast, successful rooting without applied auxin has been reported in a number of tropical tree species, such as *Shorea macrophylla* (Lo 1985), *Nauclea diderrichii* (Leakey 1990), *Milicia excelsa* (Ofori et al. 1996), *Allanblackia floribunda* (Atangana et al. 2006). Such contrasting results may be due to the variation in endogenous auxin contents at time of severance (Hartmann et al. 1990). We also think that, the reason behind the higher efficiency of rooting under low concentration of IBA for this species may be related to the eventual higher level of endogenous auxin content in the cuttings. On the other hand, higher concentrations seems to be detrimental than beneficial. We postulate that at higher concentrations IBA may have some negative impact against the naturally occurring growth hormones in the cuttings. These need to be investigated further.

The results from survival percentage of treated cuttings is indicative of some kind of toxic effect from higher concentration of IBA on the stocklings. However, the rooting percentage was higher for 0.4% IBA compared to control which means that the negative effect of high concentration of IBA for this species is more concerned with the performance and quality of the roots generated rather than the number or lengths of the roots. Future studies may clarify our understanding of such dose dependent toxicity.

No significant difference in mean root number of *L. monopetala* cuttings for different IBA concentrations was found in this study which is in agreement with Shiembo et al. (1996) for *Irvingia gabonensis*, Ofori et al. (1996) for *Milicia excelsa*, Tchoundjeu et al. (2004) for *Pausinystalia johimbe*, Baul et al. (2009) for *Stereospermum suaveolens* with different IBA concentrations and Tchoundjeu et al. (2002) for *Prunus africana* at different rooting aids like indole-3-acetic acid (IAA), IBA and 1-naphthalene acetic acid (NAA) dissolved in 10 µl of industrial alcohol. In contrast, studies on *Khaya ivorensis* (Tchoundjeu and Leakey, 1996), *Cordia alliodora* (Mese'n et al. 1997), *Juniperus*

procera (Negash, 2002), and *Dalbergia melanoxylon* (Amri et al. 2009) demonstrated the significant difference in mean root numbers owing to the variations in IBA concentrations.

The present study also showed that there was no significance difference in mean length of the longest root of cuttings of *L. monopetala* for different IBA concentrations. This is in agreement with the findings of Baul (2006) for another tropical medicinal tree species, *Holarrhena pubescens*. This might be due to the fact that all the cuttings were taken from mature mother trees causing decrease in the content of endogenous auxins in the stem cuttings which may inhibit rooting (Husen and Pal 2006; Opuni-Frimpong et al. 2008) where IBA influence was not strong on rooting ability, roots number and root length (Swamy et al. 2002; Rout 2006; Amri et al. 2009). However, the findings from studies on *Robinia pseudoacacia* and *Grewia optiva* [Swamy et al. 2002], *Tectona grandis* [Husen and Pal 2007b], and *Khaya anthotheca* and *Khaya ivorensis* [Opuni-Frimpong et al. 2008)] highlighted the significant enhancement of their mean root length for different IBA concentrations.

Our results indicated that the survival percentage of the established rooted cuttings (stocklings) after transferring into polythene bags was found the highest for the cuttings treated with 0%, 0.1% and 0.2% IBA concentration which is consistent with the observations of Minadawati and Rostawati (1989) for *Agathis loranthifolia* and Baul et al. (2009) for *Stereospermum suaveolens* at the concentration of 0.2% IBA.

There was significance difference in mean shoot number of stocklings among different concentrations of IBA in the study. This finding indicated that mean shoot number per cutting was the highest for 0.1% IBA which is in disagreement with the study on *Dalbergia sissoo* (Husen 2004) and on *Tectona grandis* (Husen and Pal 2007b) where mean shoot number increased with 0.2% and 0.4% IBA concentrations, respectively.

As wild trees grow slowly it is necessary to increase the growth by applying different concentrations of fertilizers. Plant nutrients- Nitrogen (N), Phosphorus (P) and Potassium (K) are essential for crop growth and development (Landis et al. 1989; Hassan and Leitch 2000). Regarding the impact of applied two different fertilizer treatments of three fertilizers viz. Urea, TSP (Triple super phosphate) and MOP (Muriate of potash) on growth study it was depicted that different concentrations of fertilizer had no significant positive impact on stem and collar diameter elongation, expansion of leaf area and root biomass production of the stocklings under study. These findings are in contrast to past investigations, for example, as on *Cornus florida* (Warren 1993), *Acacia mangium* (Paul and Hossain 1996), *Cedrela odorata*, *Swietenia macrophylla* (Mexal et al. 2002), *Ficus auriculata*, *Ficus hispida*, *Dillenia indica* (Islam 2005) and *Stereospermum suaveolens* (Baul et al. 2009) for growth of stocklings. Also Yeager and Wright (1981), Rieckermann et al. (1999) reported the opposite results on root biomass decrease as a result of high level of urea fertilization. In the present study, the increment of leaf area of stocklings was significantly higher under T0 (no fertilizer) compared with that under T1 (10 g of Urea+20 g of TSP (Triple super phosphate) +10 g of MOP (Muriate of potash) dissolved in 1 L of water) and T2 (10 g of

Urea+20 g of TSP (Triple super phosphate) + 10 g of MOP (Muriate of potash) dissolved in 2 L of water), while the increment of stem length, collar diameter and root biomass did not vary significantly among different levels of fertilizer. In case of stem and collar diameter elongation, T0 and T1 respectively showed better results with compared to other fertilizer treatments. This may be due to the influence of different IBA concentrations the cuttings were subjected to, which has not been considered in designing the growth performance study.

From our results, we can conclude that the rooting percentage, stecklings' survival percentages and subsequent growth parameters are all indicative of the the amenability of *L. monopetala* to vegetative propagation without IBA or with low concentration (for example 0.1% IBA) IBA treatment of leafy branch cutting. We have not studied other hormones like IAA or NAA which can be investigated by further research involving the species. Moreover, the application of fertilizer would appear to be unnecessary for the growth of stecklings in polythene bags. Whether other combinations of fertilizers may have more positive effects or not is yet to be investigated. Through this research, it has become clear that, vegetative propagation can be a very easy, cheap and effective mean in producing the planting stock of desirable attributes for the domestication of the species to supply enough medicinal raw materials to the rural communities and pharmaceutical companies. Furthermore, the present work opens a new window of opportunity both for researchers and farmers for further research in genetic improvement and large scale cultivation, sustainable utilization and conservation of *L. monopetala* using vegetative propagation techniques.

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